PARACEST detection in-vivo using WALTZ-16

E. Vinogradov[1], H. He[2], A. Lubag[3], V. Humblet[4], J. A. Balschi[2], A. D. Sherry[3,5], J. V. Frangioni[4], R. E. Lenkinski[1]

[1]Department of Radiology, Beth Israel Deaconess Medical Center, Boston, MA, United States, [2]NMR Laboratory for Physiological Chemistry, Brigham and Woman's Hospital, Boston, MA, United States, [3]Department of Radiology, University of Texas Southwestern Medical Center, Dallas, TX, United States, [4]Department of Hematology/Oncology, Beth Israel Deaconess Medical Center, Boston, MA, United States, [5]Department of Chemistry, University of Texas at Dallas, Dallas, TX, United States

Introduction

Chemical Exchange Saturation Transfer (CEST) imaging[1] detects exchange mediated saturation transfer between bound and bulk protons. CEST image contrast can be turned "on" and "off" via RF pre-saturation (CW irradiation). As chemical exchange can be quite sensitive to the contrast reagent environment, the CEST effect can be used to image physiological parameters such as pH[1-5]. Exogenous paramagnetic lanthanide complexes (PARACEST) exhibit slow exchange kinetics and large chemical shifts for the lanthanide-bound water molecule[6]. CEST imaging can potentially detect μM PARACEST concentrations [PARACEST]; however, achieving the maximal effect for a [PARACEST] may require CW irradiation that is above FDA guidelines[7] for human imaging. Another potential complication with the standard CW pre-saturation is that the frequency of the bound water peak needs to be known *a-priori*; this frequency may vary with temperature and thus be problematic *in-vivo*. We developed an alternate approach using exchange- and relaxation- sensitive RF pulses applied at the bulk water frequency. This pulse sequence used a WALTZ-16 pulse train[8] in which the 90^0 pulses were replaced by 2.5 msec pulses (WALTZ-16*); total RF pulse train time of 220 msec. At the end of the WALTZ-16* pulse, non-exchanging water spins return to the Z axis. The exchanging protons "leak away" during RF application and, hence, do not experience a full 360^0 rotation. The resultant Z magnetization thus decreases. *In-vitro* studies have shown that the WALTZ-16* sequence with an RF intensity of 200 Hz can detect (Tm,Dy) [PARACEST] as low as a few tens of μM[9]. Here we report the initial use of the WALTZ-16* sequence to detect PARACEST effects *in-vivo*.

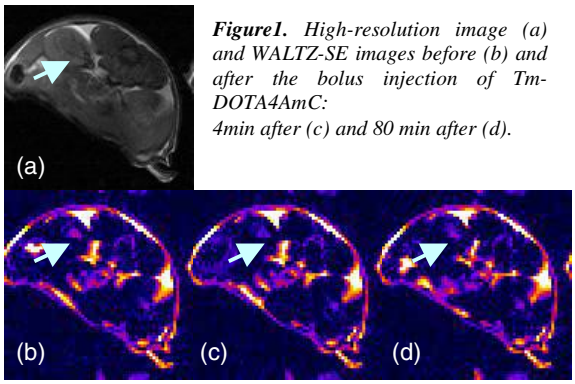

Figure1. *High-resolution image (a) and WALTZ-SE images before (b) and after the bolus injection of Tm-DOTA4AmC: 4min after (c) and 80 min after (d).*

Methods

Experiments were performed on a Varian Unity INOVA 400MHz vertical wide bore or 200MHz horizontal wide bore systems with volume imaging coils. Male C57BL6 mice (22-29g) were anesthetized by pentobarbitol injection (65mg/kg bw) and maintained with inhaled isoflurane. Bolus injections of TmDOTA-4AmC PARACEST were used in all the experiments. The contrast agent was delivered via tail vein injection in 200 MHz studies and via the jugular vein in 400 MHz studies. Temperature, ECG and breathing rate were monitored and within normal range. The total injected volume was 100 -150μL at [PARACEST] = 10-40 mM.

Axial image slices included at least one kidney. Spin echo images with WALTZ-16* before imaging pulses (WALTZ-SE) were acquired with the following parameters: WALTZ-16* B_1 range ~ 200Hz, TR 1s, TE 8.2 - 8.8 msec, FOV 25 cm, matrix size 64 x 64, slice thickness 2 mm, 4 averages. Total scan time was 4 min2 sec. Dynamic studies were performed by acquiring WALTZ-SE images continuously for 1-3 hours after the bolus injection. ROI were hand-drawn in the kidneys. Mean ROI intensity was monitored as a function of time.

Results

To verify that the WALTZ-16* sequence is able to create detectable contrast differences *in-vivo*, WALTZ-SE images were acquired before and after PARACEST injection. Figure 1 shows an axial high-resolution image (a)of the kidney, and WALTZ-SE images acquired immediately before(b), 4min(c) after and 80min(d) after the beginning of the bolus. The PARACEST contrast manifests itself as a darkening of the image that is clearly visible. To monitor the dynamics and quantify the effect size, the hand-drawn ROIs were placed over the cortex and medulla. The measured mean ROI intensities (**I**) were normalized by division by the **I** measured right before the injection (see Fig 2). Figure 2 displays the changes in normalized **I** after PARACEST injection. At 40mM [PARACEST] bolus effect sizes of up to 70% were measured. At the lowest [PARACEST] used so far, 10mM, the effect size was about 50%.

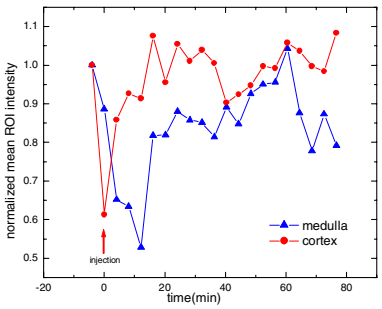

Figure 2. *Normalized mean ROI for cortex and medulla. Normalized using mean ROI prior to injection (t=-4min)*

Discussion

The WALTZ sequence can be used for the visualization of PARACEST reagents *in-vivo*. A limitation of the sequence is the signal decrease due to the direct saturation. However, the residual signal is sufficient to detect changes in the contrast. SNR should be incremented in order to shorten detection time and allow detection of microMolar concentrations. Other on-resonance sequences of shorter length, which may have similar capabilities as WALTZ-16, might be superior. We are exploring ways of utilizing them. Despite the direct saturation drawback, the effect sizes created are big, and the [PARACEST] detected are by much greater than the limiting concentration. In the following experiments the concentrations will be further reduced until the low limit will be found. Additional studies of the physiological dynamics of the contrast agent are underway using a Gd derivative to provide the rates of renal clearance. To date, a PARACEST Eu complex, which has slower exchange dynamics than Tm or Dy complexes, was used in two mice, with CW saturation (1sec length 1kHz intensity). The animal temperature increased by several of degrees, and the frequency of bound-water peak shifted by more than a kHz. This suggests that shorter length broad-band saturation needs to be used to fully exploit the potential of such PARACEST with slower exchange dynamics than Tm or Dy.

Conclusion

WALTZ-SE successfully imaged PARACEST effects *in-vivo*. The advantages of WALTZ-16* are low RF power deposition and no need in the *a priori* knowledge of the exact bound water peak frequency. Signal reduction due to direct saturation is a disadvantage. Work is in progress to further improve this sequence in terms of SNR and verify the [PARACEST] low-detection limit *in vivo*. We have developed quantitative formulas for the dependence of the effect sizes on system parameters and [PARACEST] and are currently validating their accuracy.

References

1.K Ward *et.al. Magn.Reson.Med.* 44(2000)799; 2. J Zhou *et.al. Nat.Med.* 9(2003) 1085; 3. S.Aime *et.al. Magn.Reson.Med.* 47(2002)639; 4. S Zhang *et.al. Angew Chem Int Ed* 41(2002) 1919; 5. S.Aime *et.al. Angew Chem Int Ed*41(2002) 4334; 6. S Zhang *et al J Am Chem Soc* 124(2002)4226; 7**.** S.Zhang, *et.al., Acc.Chem.Res.*;
8. A.J.Shaka, *et.al., J.Magn.Reson.,* 53(1982)313; 9. E.Vinogradov *et.al. J Magn Reson* 176(2005)54.